Hence, the amendments to the specification and claims, and the addition of the Abstract and Sequence Listing do not constitute new matter.

The Examiner is invited to contact the undersigned at his Washington telephone number on any questions which might arise.

Respectfully submitted,

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A P P E N D I X

Marked-Up Version of Amendments

IN THE SPECIFICATION:

The specification is amended as follows:

Page 1, before line 3,

-- This Application is a Continuation of PCT/GB99/04417 (published under PCT Article 21(2) in English) filed December 23, 1999. -- is inserted.

Page 9, lines 27-37 and Page 10, lines 1-5 are changed as follows:

In an exemplary procedure, all target molecules that do not end with AAAA (SEQ ID NO:1) (when the probe ends in TTTT (SEQ ID NO:2)) would not bind and would be removed. Similarly at other addresses, target molecules having particular end sequences would bind selectively. The target molecules may be double stranded (with single stranded overhangs) or single stranded such that sequences could be bound to and identified at the terminal ends or also internally, respectively. If PNA was used as the complementary probe, since such molecules are able to bind to double stranded forms, internal sequences of double stranded forms could also be bound. In general terms this technique is referred to herein as sorting based on one or several end base pairs and may be performed in one or more cycles. This technique may be coupled to other techniques as described herein.

Page 13, lines 20-37 and Page 14, line 1 are changed as follows:

In general terms an example of the process may be described as follows. Base pairs in the target nucleic acid material are

associated with four different tags (hereafter called magnifying tags) that represent each of the four bases Adenine, Cytosine, Guanine, and Thymine. Thus, where there was an A-T base pair "magnifying tag A" is associated, C-G is associated with "magnifying tag C", etc. Thereby new DNA molecules are generated where the original base order of e.g. ACGTT (SEQ ID NO:3) is augmented by "magnifying tag A" - "magnifying tag C" - "magnifying tag G", etc. Each magnifying tag provides a means of producing a signal and may in a preferred feature be a polynucleotide molecule. In that case the length of the four tags may vary from two base pairs to several hundred kbp (or more if desired), according to requirements. Correspondingly, the DNA fragments can contain reporter genes and other biological information or consist only of sequences without a known biological function.

Page 20, lines 33-37 and Page 21, lines 1-7 are changed as follows:

Appropriate adapters may then be used to bind to, and thereby allow magnification of, one or more bases of the overhang. In the case of single base magnification, degenerate adapters having single stranded portions of the form, e.g. for a four base overhang, ANNN (SEQ ID NO:4), TNNN (SEQ ID NO:5), CNNN (SEQ ID NO:6) and GNNN (SEQ ID NO:7) and magnifying tags A, T, C and G, respectively may be used. Alternatively the adapters may carry more than one magnifying tags corresponding to more than one of the overhang bases, e.g. having an overhang of ATGC (SEQ ID NO:17), with corresponding magnifying tags to one or more of those bases attached in linear fashion where appropriate.

Page 34, lines 5-19 are changed as follows:

The DNA molecules in well 1 have AAAA (SEQ ID NO:1) overhangs, while the DNA molecules in well 2 have AAAC (SEQ ID NO:8) overhangs, etc. The 256 wells thereby cover all permutations of overhangs on four bases. When the target DNA are added to the wells together with ligase, the DNA molecules with TTTT (SEQ ID NO:2) overhangs will attach themselves to well 1, the target DNA with TTTG (SEQ ID NO:9) overhangs to well 2, etc. After having washed off target DNA molecules that were not ligated to the sorting adapters, IIS enzyme is added so that the target DNA molecules are freed at the same time as a new overhang is created that represents the next four base pairs in the target sequence. This overhang can then be used as the starting point for a new round of sorting, or one may proceed with conversion/magnification.

Page 34, lines 27-33 are changed as follows:

Instead of using different wells, an alternative would be to use different positions on a "microarray". At address 1 it is only DNA molecules that end with TTTT (SEQ ID NO:2) that are fixed, at address 2 it is DNA molecules with TTTG (SEQ ID NO:9) ends that are fixed, etc. Other alternatives are to let DNA molecules with different ends attach/convert at different times, the use of gel sorting, etc.

Page 34, lines 34-37 and Page 35, lines 1-11 are changed as follows:

For example, one may use a strategy where there are 256 different sorting adapters distributed among 256 squares on a "microarray". In square 1, there are sorting adapters with AAAA (SEQ ID NO:1) overhangs, in square 2, they have AAAC (SEQ ID NO:8) overhangs, etc. Thus, the target DNA molecules will be

sorted so that those with TTTT (SEQ ID NO:2) overhangs are attached to square 1, GTTT (SEQ ID NO:10) overhangs to square 2, etc. By also fixing the other end of the DNA piece to the substrate, e.g. with biotin/streptavidin, one can then continue to the next conversion/magnification step without the DNA molecules leaving their position on the reading plate. Another strategy for preventing the DNA molecules from leaving their positions is to use a reading plate that is divided into 256 wells/spaces.

Page 56, lines 25-31 are changed as follows:

If sorting is used, it is possible to sort the same piece of sequence several times. For example, all target DNA that begin with AAAA (SEQ ID NO:1) are sorted into well 1. Then the same procedure is repeated where incorrectly sorted DNA molecules that do not end with AAAA (SEQ ID NO:1) are washed away. The procedure can in principle be repeated until the desired error percentage is obtained.

Page 61, lines 19-24 are changed as follows:

Figure 24 shows the sorting method described herein which is performed on a microarray in which overhangs of 4 bases in the target DNA are mixed with a microarray with 256 addresses and ligated. Address 1 contain AAAA (SEQ ID NO:1) overhangs and thus binds to target molecules with TTTT (SEQ ID NO:2) overhangs; and

Page 62, lines 11-25 are changed as follows:

2. Base pairs in the DNA pieces are replaced with four different DNA sequences (hereafter called DNA fragments, corresponding to the magnifying tags) that represent each of the four bases Adenine, Cytosine, Guanine, and Thymine. Thus, where there was an A-T base pair "fragment A" is inserted, C-G is

replaced by "fragment C", etc. Thereby new DNA molecules are generated where the original base order of e.g. ACGTT (SEQ ID NO:3) is replaced by fragment A - fragment C - fragment G, etc. The length of these four DNA fragments can, in principle, vary from two base pairs to several hundred kbp (or more if desired), according to requirements. Correspondingly, the DNA fragments can contain reporter genes and other biological information or consist only of sequences without a known biological function.

Page 77, lines 30-37 and Page 78, lines 1-4 are changed as follows:

Since the overhangs with target DNA in the above-mentioned example seek complementary overhang, each converted DNA piece will be hybridized/ligated to encountered DNA pieces. This creates a chain of magnifying tags (signal chain) that provides information about sequence pieces of 8 base pairs interrupted by 22 unknown bases (e.g. AGCTGTGA N22 AGTCTGCA N22 TGAC (SEQ ID NO:11)). The number of unknown base pairs is determined by the initial length of the DNA piece minus the number of base pairs converted per DNA piece. Based on overlaps between signal chains, it is then possible to reconstruct the target sequence even in areas with repetitive sequences.

Page 80, lines 31-37 and Page 81, line 1 are changed as follows:

Methods

1) The starting point is a scanning surface consisting of 65,536 addresses. A perpendicular anchoring line with single-stranded octamers is attached to each address. AAAAAAAA (SEQ ID NO:12) octamers are anchored to the plate at address 1, AAAAAAAC (SEQ ID NO:13) octamers to the plate at

[address 2,etc,] address 2, etc, so that all the 65,536 octamer permutations each have their own address.

Page 83, lines 10-20 are changed as follows:

7) The target DNA is distributed from the 256 wells between 256 microarrays as described in Example 12. All the microarrays are alike and consist of 256 addresses with sorting adaptors with 4-base overhangs that can complement the overhangs made in step 6). At address 1 the sorting adaptors have AAAA (SEQ ID NO:1) overhangs, at address 2 they have AAAC (SEQ ID NO:8) overhangs etc.

8) Ligase is added and the mixture incubated. At address 1 there will be target DNA with TTTT (SEQ ID NO:2) overhangs, at address 2 there will be target DNA with TTTG (SEQ ID NO:9) overhangs, etc.

Page 84, lines 14-26 are changed as follows:

Results

The presence or absence and size of molecules at particular addresses indicates both sequence information and its position. Thus if address 1 of microarray 1 contains DNA molecules of [100micrometers] 100 micrometers, this indicates that the sequence corresponding to the octamer used (albeit by 2-step sorting) to bind that molecule is present at +200kb (e.g. TTTTTTTT (SEQ ID NO:14)). Similar the presence of 2 differently sized molecules would indicate a repeat of the particular sequence. The absence of any molecules at a particular address would indicate the absence of the sequence complementary to the immobilizing octamer in the target sequence.

Page 94, lines 26-30 are changed as follows:

Method

1) The DNA molecule that is to be sequenced, ACGTGAGCT (SEQ ID NO:15) is fixed with one end to a streptavidin-covered plate. The fixing mechanism should be a mechanism other than streptavidin/biotin.

Page 95, lines 1-6 are changed as follows:

4) A solution with various adapters and ligases is then added. Figure 19 shows an adapter that has recognized and bound to the ACGT (SEQ ID NO:16) overhang. In addition to fragments labelled with fluorescence that correspond to the ACGT (SEQ ID NO:16) overhang, two or more biotin molecules have been incorporated on the adapter.

Page 96, lines 36-37 and Page 97, lines 1-8 are changed as follows:

Method

A DNA chip is used which is divided into 256 addresses. Each address contains sorting adaptors, an overhang, a binding site for a class IIS restriction endonuclease and a binding site for a restriction endonuclease that makes a blunt end cut. The overhangs vary from address to address so that address 1 has sorting adaptors with an AAAA (SEQ ID NO:1) overhang, address 2 has an AAAC (SEQ ID NO:8) overhang, etc. In addition all the addresses are covered with a molecule with binding properties, e.g. streptavidin.

Page 97, lines 13-18 are changed as follows:

2) The fragments are introduced to the solid support carrying the sorting adaptors to which they are ligated. DNA pieces with a TTTT (SEQ ID NO:2) overhang will ligate to address 1 in which the sorting adaptors have the complementary overhang AAAA (SEQ

ID NO:1), DNA pieces with a GTTT (SEQ ID NO:10) overhang will ligate to address 2 etc.

IN THE CLAIMS:

Claim 1-25 are being cancelled.

New Claims 26-64 are being added.

IN THE ABSTRACT:

An Abstract is added.

SEQUENCE LISTING:

A Sequence Listing is added.